PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : C07D 295/10, A61K 31/495 — A1

(11) International Publication Number: WO 99/06382

(43) International Publication Date: 11 February 1999 (11.02.99)

(21) International Application Number: PCT/EP98/04796

(22) International Filing Date: 31 July 1998 (31.07.98)

(30) Priority Data:
MI97A001862 1 August 1997 (01.08.97) IT
MI97A001863 1 August 1997 (01.08.97) IT

(71) Applicant *(for all designated States except IT)*: RECORDATI S.A., CHEMICAL AND PHARMACEUTICAL COMPANY [CH/CH]; Piazzo Boffalora, 4, CH-6830 Chiasso (CH).

(71) Applicant *(for IT only)*: RECORDATI INDUSTRIA CHIMICA E FARMACEUTICA S.P.A. [IT/IT]; Via M Civitali, 1, I-20148 Milano (IT).

(72) Inventors: LEONARDI, Amedeo; Via A. Poliziano, 16, I-20154 Milano (IT). MOTTA, Gianni; Via Ungaretti, 10, I-20030 Barlassina (IT). RIVA, Carlo; Via Walder, 10, I-21100 Varese (IT). GUARNERI, Luciano; Via Canova, 18, I-20024 Garbagnate Milanese (IT). POGGESI, Elena; Via Ricciarelli, 37, I-20148 Milano (IT).

(74) Agent: SERJEANTS; 25 The Crescent, King Street, Leicester LE1 6RX (GB).

(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published

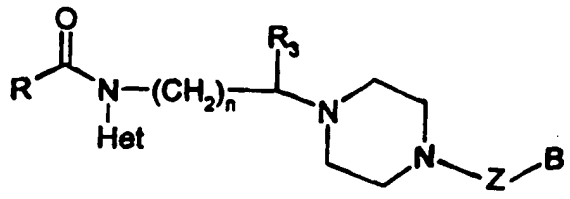
With international search report.

Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

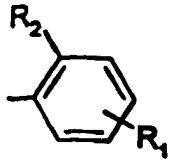
(54) Title: 1,4-DISUBSTITUTED PIPERAZINES

(57) Abstract

Compounds of formula (I) (n = 1 or 2, Het = monocyclic heteroaryl, R = cycloalkyl or monocyclic heteroaryl, R_3 = H or lower alkyl, Z = bond, $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$, B = substituted or unsubstituted aryl or heteroaryl) bind to 5HT1A receptors and are therefore useful for the treatment of neuromuscular dysfunctions of the lower urinary tract. The compounds in which Z = bond, B = a substituted phenyl group of formula (a) (R_1 = H, halogen, alkoxy, NO_2, NH_2, NH(acyl), $NHSO_2$(alkyl), R_2 = halogen, alkoxy, polyfluoroalkoxy, CN, $CONH_2$; provided that if R_1 NH(acyl) or $NHSO_2$(alkyl) then R_2 = polyfluoroalkoxy) and the compounds in which Z = $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$ are claimed per se; other compounds are claimed for use in preparation of medicaments for treating neuromuscular dysfunctions of the lower urinary tract.



(I)



(a)

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

1,4-Disubstituted Piperazines

Field of the Invention

This invention relates to novel 1,4-disubstituted piperazines that bind to serotonergic receptors, to pharmaceutical compositions containing them, and to uses for such derivatives and compositions.

Background Of The Invention

In mammals, micturition (urination) is a complex process that requires the integrated actions of the bladder, its internal and external sphincters, the musculature of the pelvic floor, and neurological control over these muscles at three levels (in the bladder wall or sphincter itself, in the autonomic centres of the spinal cord, and in the central nervous system at the level of the pontine micturition centre (PMC) in the brainstem (pons) under the control of cerebral cortex) (De Groat, *Neurobiology of Incontinence*, (Ciba Foundation Symposium 151:27, 1990). Micturition results from contraction of the detrusor muscle, which consists of interlacing smooth muscle fibres under parasympathetic autonomic control from the sacral spinal cord. A simple voiding reflex is formed by sensory nerves for pain, temperature, and distension that run from the bladder to the sacral cord. However, sensory tracts from the bladder also reach the PMC, resulting in the generation of nerve impulses that normally suppress the sacral spinal reflex arc controlling bladder emptying. Thus, normal micturition is initiated by voluntary suppression of cortical inhibition of the reflex arc and by relaxation of the muscles of the pelvic floor and the external sphincter. Finally, the detrusor muscle contracts and voiding occurs.

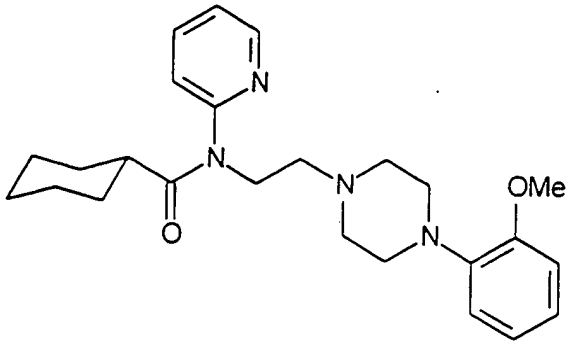
Abnormalities of lower urinary tract function, e.g., dysuria, incontinence, and enuresis, are common in the general population. Dysuria includes urinary frequency, nocturia, and urgency, and may be caused by cystitis, prostatitis or benign prostatic hypertrophy (BPH) (which affects about 70% of elderly males), or by neurological disorders. Incontinence syndromes include stress incontinence, urgency incontinence, and overflow incontinence. Enuresis refers to the involuntary passage of urine at night or during sleep.

Prior to the work of the present inventors, treatment of neuromuscular dysfunction of the lower urinary tract has involved administration of compounds that act directly on the bladder muscles, such as flavoxate, a spasmolytic drug (Ruffman, *J. Int.Med.Res.* 16:317, 1988) also active on the PMC (Guarneri et al., *Drugs of Today* 30:91, 1994), or anticholinergic compounds such as oxybutynin (Andersson, *Drugs* 35:477, 1988). The use of a1-adrenergic receptor antagonists for the treatment of BPH is also common but is based on a different mechanism of action. (Lepor, *Urology*, 42:483, 1993).

However, treatments that involve direct inhibition of the pelvic musculature (including the detrusor muscle) may have unwanted side effects such as incomplete voiding or

accommodation paralysis, tachycardia and dry mouth (Andersson, *Drugs* 35:477, 1988). Thus, it would be preferable to utilise compounds that act via the peripheral or central nervous system to, for example, affect the sacral spinal reflex arc and/or the PMC inhibition pathways in a manner that restores normal functioning of the micturition mechanism.

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-methoxyphenyl)-piperazine (Compound A below) is described in GB 2255 337 A and is reported to possess 5-HT_{1A} antagonistic properties. It is also disclosed that it can be used for the treatment of central nervous system disorders, for example as an anxiolytic agent in the treatment of anxiety.

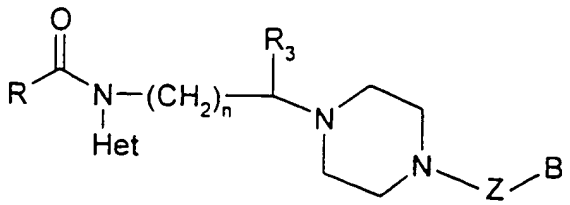


(A)

The compounds of the present invention, described below, are structurally different from compound A because of the novel substitutions on the aromatic ring bound to the piperazine moiety and of the insertion of a series of spacers (Z) between the piperazine and phenyl rings. These structural variations are not disclosed by GB 2255337 A, particularly with regard to compounds that can be used to improve urinary tract function. These novel compounds also have a longer duration of action than does A in pharmacological tests predictive of activity on the lower urinary tract. This is especially true with respect to the activity of the compounds of the invention against urinary incontinence, which is a novel therapeutic indication for this class of compounds acting at the 5-HT_{1A} receptor.

SUMMARY OF THE INVENTION

In one aspect, the invention relates to the use of compounds of formula I:



(I)

wherein

n is 1 or 2.

Het represents a monocyclic heteroaryl group,

R represents a cycloalkyl or a monocyclic heteroaryl group,

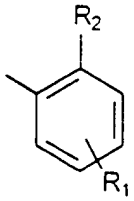
R_3 represents a hydrogen atom or a lower alkyl group,

Z represents a bond or a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$, each of which is depicted with its left end being the end which attaches to the piperazine ring and its right end being the end which attaches to group B, and

B represents a substituted or unsubstituted aryl or heteroaryl radical,

for the preparation of a medicament for the treatment of neuromuscular dysfunction of the lower urinary tract in a mammal.

The compounds I in which Z represents a bond, B represents a substituted phenyl group of the formula



wherein

R_1 represents a hydrogen or halogen atom or an alkoxy, nitro, amino, acylamino or alkylsulphonylamino group, and

R_2 represents a halogen atom or an alkoxy, polyfluoroalkoxy, cyano or carbamoyl group, but

if R_1 does not represent an acylamino or alkylsulphonylamino group, then R_2 represents a polyfluoroalkoxy group;

and the compounds I in which Z represents a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$ are new and are provided in another aspect of the invention.

The invention also includes the enantiomers, diastereomers, N-oxides, crystalline forms, hydrates and pharmaceutically acceptable salts of these compounds, as well as metabolites of these compounds having the same type of activity (hereafter sometimes referred to as "active metabolites").

The invention further provides pharmaceutical compositions comprising a compound of formula I or an enantiomer, diastereomer, N-oxide, crystalline form, hydrate or pharmaceutically acceptable salt of the compound, in admixture with a pharmaceutically acceptable diluent or carrier.

The compounds of the invention are useful for reducing the frequency of bladder contractions due to bladder distension by administering one or more selected compounds of Formula I to a mammal (including a human) in need of such treatment, in an amount or amounts effective for the particular use.

The compounds of the invention are also useful for treating disorders of the urinary tract in a subject in need of such treatment, by administering an amount of a compound of Formula I effective to ameliorate at least one of urinary urgency, increased urinary frequency, incontinence, urine leakage, enuresis, dysuria, urinary hesitancy, and difficulty in emptying the bladder.

The compounds of the invention have been found to bind to 5-HT_{1A} serotonergic receptors and, by virtue of this activity, may be found useful for the treatment of CNS disorders due to serotonergic dysfunction. Such dysfunctions include anxiety, depression, hypertension, sleep/wake cycle disorders, feeding behaviour, sexual function and cognition disorders in mammals, particularly in humans. Treatment may be effected by delivering to the environment of the 5-HT_{1A} serotonergic receptors, e.g., to the extracellular medium (or by administering to a mammal possessing such receptors), an effective amount of a compound of the invention.

DETAILED DESCRIPTION OF THE INVENTION

All patents, patent applications, and literature references cited in the specification are hereby incorporated by reference in their entirety. In the case of inconsistencies, the present disclosure, including definitions, will prevail.

The activity of the compounds of the invention as inhibitors of frequency of micturition renders them useful for the treatment of neuromuscular dysfunctions of the lower urinary tract in mammals, including without limitation dysuria, incontinence and enuresis.

Surprisingly, the introduction of selected substituents on the phenyl ring directly bound to the piperazine moiety of formula I and of the Z group in compounds of formula I impart to these compounds a distinctly longer duration of action than is possessed by compound A.

Preferred cycloalkyl groups R are C_5-C_7 cycloalkyl groups; preferred monocyclic heteroaryl groups R and Het are those having 5 to 7 ring atoms including one or more hetero atoms (e.g. oxygen, nitrogen, or sulphur). Lower alkyl, as used herein, includes C_1 to C_6 alkyl. Alkyl, when used in, for example "alkylsulphonylamino", also means lower alkyl, preferably C_1 to C_6 alkyl.

Preferred heteroaryl groups B are mono or bicyclic aromatic radical having from 5 to 12 ring atoms including one or more heteroatoms (e.g. nitrogen, oxygen, sulphur).

Preferably n is 1, R represents a cyclohexyl group, Het represents a 2-pyridyl group and R_3 represents a hydrogen atom, all independently of each other. A preferred group of compounds is that in which Z represents a bond. R_1 represents a hydrogen or halogen atom

or a nitro, amino, acylamino or alkylsulphonylamino group, and R_2 represents an alkoxy, trifluoroalkoxy, cyano or carbamoyl group. More preferred is the group of compounds in which Z represents a bond and B represents a 2-trifluoromethoxyphenyl, 2-(2,2,2-trifluoroethoxy)-phenyl, 5-chloro-2-(2,2,2-trifluoroethoxy)-phenyl, 4-acetamido-2-methoxyphenyl, 2-methoxy-4-methylsulphonylamino-phenyl, 2-methoxy-4-pivaloylaminophenyl or 4-butanoylamino-2-methoxyphenyl group.

When Z does not represent a bond, B preferably represents an unsubstituted phenyl group or a substituted phenyl group, the substituents being selected from halogen atoms and alkoxy, cyano, nitro, amino, acylamino, alkylsulphonylamino, polyfluoroalkoxy and carbamoyl groups. Another preferred group of compounds is that in which Z represents a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$ or $-C(O)-$. A still further preferred group of compounds is that in which Z does not represent a valence bond and in which B represents a phenyl, 2,5-dichlorophenyl or 2-bromo-5-methoxyphenyl group.

Subjects who can benefit from administration of the compounds and compositions of the invention include humans who are affected by neuromuscular dysfunction of the lower urinary tract, described by E.J. McGuire in "Campbell's UROLOGY" 5th Ed. 616-638, 1986, W.B. Saunders Company, and also include patients affected by any physiological dysfunction related to impairment of 5-HT_{1A} receptor function. Such dysfunctions include, without limitation, central nervous system disorders such as depression, anxiety, eating disorders, sexual dysfunction, addiction, and related problems.

The present invention encompasses pharmaceutical formulations comprising the compounds disclosed above, as well as methods employing these formulations for treating neuromuscular dysfunction of the lower urinary tract such as dysuria, incontinence, enuresis, and the like. Dysuria includes urinary frequency, nocturia, urgency, and difficulty in emptying the bladder, i.e., a suboptimal volume of urine is expelled during micturition.

Incontinence syndromes include stress incontinence, urgency incontinence, and overflow incontinence. Enuresis refers to the involuntary passage of urine at night or during sleep.

Without wishing to be bound by theory, it is believed that administration of 5-HT_{1A} receptor antagonists prevents unwanted activity of the sacral reflex arc and/or cortical mechanisms that control micturition. Thus it is contemplated that a wide range of neuromuscular dysfunctions of the lower urinary tract can be treated using the compounds of the present invention.

An "effective amount" of the compound for treating a urinary disorder is an amount that results in measurable amelioration of at least one symptom or parameter of the disorders described above.

An effective amount for treating the disorder can easily be determined by empirical methods known to those of ordinary skill in the art, such as by establishing a matrix of dosages and frequencies of administration and comparing a group of experimental units or subjects to each point in the matrix. The exact amount to be administered to a patient will vary depending on the state and severity of the disorder and the physical condition of the patient. A measurable amelioration of any symptom or parameter can be determined by a physician skilled in the art or reported by the patient to the physician. It will be understood that any clinically or statistically significant attenuation or amelioration of any symptom or parameter of urinary tract disorders is within the scope of the invention. Clinically significant attenuation or amelioration means perceptible to the patient and/or to the physician.

For example, a single patient may suffer from several symptoms of dysuria simultaneously, such as, for example, urgency and excessive frequency of urination, either or both of which may be reduced using the methods of the present invention. In the case of incontinence, any reduction in the frequency or volume of unwanted passage of urine is considered a beneficial effect of the present methods of treatment.

The compounds of the present invention may be formulated into liquid dosage forms with a physiologically acceptable carrier, such as, for example, phosphate buffered saline or deionized water. The pharmaceutical formulation may also contain excipients, including preservatives and stabilisers, that are well-known in the art. The compounds can be formulated into solid oral or non-oral dosage units such as, for example, tablets, capsules, powders, and suppositories, and may additionally include excipients, including without limitation lubricant(s), plasticizer(s), colorant(s), absorption enhancer(s), bactericide(s), and the like.

Modes of administration include oral and enteral, intravenous, intramuscular, subcutaneous, transdermal, transmucosal (including rectal and buccal), and by-inhalation routes. Preferably, an oral or transdermal route is used (i.e., via solid or liquid oral formulations, or skin patches, respectively).

The amount of the agent to be administered can range from between about 0.01 and about 25 mg/kg/day, preferably from between about 0.1 and about 10 mg/kg/day and most preferably from between about 0.2 and about 5 mg/kg/day. It will be understood that the pharmaceutical formulations of the present invention need not in themselves contain the entire amount of the agent that is effective in treating the disorder, as such effective amounts can be reached by administration of a plurality of doses of such pharmaceutical formulations.

In a preferred embodiment of the present invention, compounds are formulated in capsules or tablets, each preferably containing 50-200 mg of the compounds of the invention, and are most preferably administered to a patient at a total daily dose of 50-400 mg, preferably

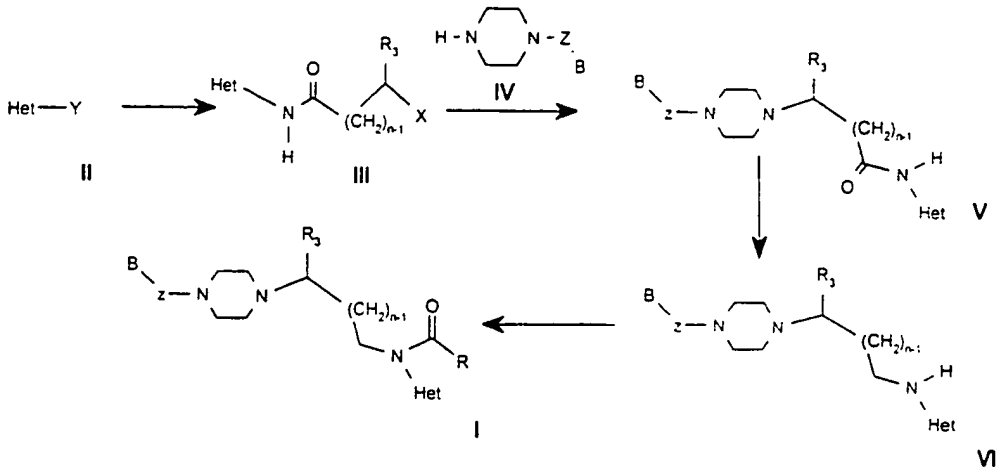
150-250 mg, and most preferably about 200 mg for relief of urinary incontinence and dysfunctions amenable to treatment with 5-HT_{1A} receptor ligands.

The methods, tables and examples provided below are intended to more fully describe preferred embodiments of the invention and to demonstrate its advantages and applicability, without in any way limiting the scope of the invention.

SYNTHESIS OF THE COMPOUNDS OF THE INVENTION

The compounds of the invention, i.e., compounds of formula (**I**), may generally be prepared as shown by Scheme 1 where Het, n, Z, B, R_3 have the same meanings as above:

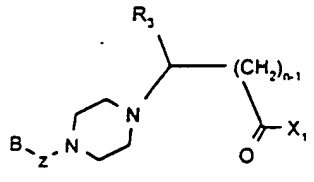
Scheme 1



The amino-substituted heteroaromatic starting materials of formula **II** ($Y=NH_2$) are commercially available, or their syntheses are well-known to those of ordinary skill in the art. The intermediates of formula **II** can be converted into intermediates with formula **III** by conventional acylation procedures known to those skilled in the art, e.g., by using acylating reagents of the formula $X\text{-}CH(R_3)\text{-}(CH_2)_{n-1}\text{-}C(O)\text{-}X_1$, where X is a leaving group, *e.g.* Br, Cl, I, p-toluenesulphonyloxy, methylsulphonyloxy and X_1, for example, is Br, Cl, OH, and the like. Where n is 2, the acylating reagent can be an acryloyl acylating reagent, which affords the 2,3-unsaturated amides of formula $Het\text{-}NH\text{-}C(O)\text{-}CH{=}CH\text{-}R_3$.

Intermediates with formula **III** can be condensed by conventional methods with the piperazine derivatives **IV**, in the presence of a base, to afford Intermediates with formula **V**.

An alternative method of preparing Intermediates of formula **V** consists of acylating starting materials of formula **II** with compounds of the formula **VII**, below, where X_1, Z, B, R_3 and n are defined as set forth above:

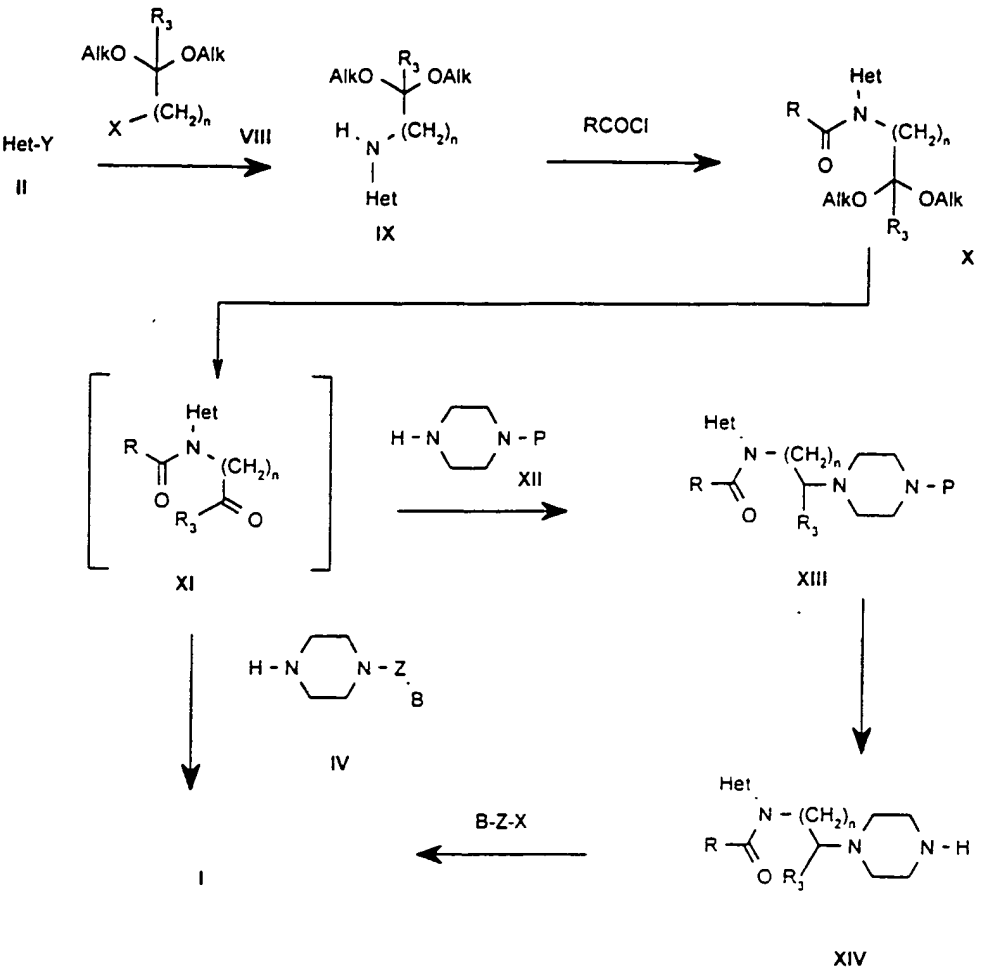


VII

The acylation can be performed by conventional procedures, known to those of ordinary skill. For example, if X_1 of the acylating agent is OH, the amine-substituted heteroaromatic can be acylated by the addition of a coupling agent (*e.g.* diethyl cyanophosphonate, dicyclohexylcarbodiimide or N,N'-carbonyldiimidazole) optionally in the presence of a promoting agent (*e.g.* N-hydroxysuccinimide, 4-dimethylaminopyridine) in an aprotic or a chlorinated solvent (*e.g.* N,N-dimethylformamide, chloroform, methylene chloride) at -20°C/140°C (Albertson, *Org. React.* **1962**, *12*, 205-218; Doherty et al., *J. Med. Chem* **1992**, *35*, 2; Staab et al., *Newer Methods Prep. Org. Chem.* **1968**, *5*, 61; Ishihara, *Chem. Pharm. Bull.* **1991**, *39*, 3236).

Other well-known reaction procedures for acylation of amines include the mixed anhydride method by reaction of intermediates with formula **VII** with an alkyl chloroformate in the presence of a tertiary amine (e.g. triethylamine) followed by addition of the heteroarylamine reagent in an aprotic solvent (e.g. dioxane, methylene chloride), optionally in the presence of, *e.g.* 1-hydroxypiperidine as a promoting agent (*Org. React.* **1962**, *12*, 157). If X_1 of the acylating agent is Cl or Br, intermediates with formula **VII** can be condensed with starting materials of formula **II** in an aprotic solvent (*e.g.* chloroform, 1,2-dichloroethane, dimethylformamide, dioxane, toluene) in the presence of a base (*e.g.* triethylamine, pyridine, 4-dimethylaminopyridine, potassium or caesium or sodium carbonate). Intermediates of formula **VII** can be synthesised from arylpiperazines and a compound of the formula $X\text{-}CH(R_3)\text{-}(CH_2)_{n-1}\text{-}C(O)\text{-}X_1$, where X is as defined above and X_1 is OH or OAlk (Alk preferably represents lower alkyl, *e.g.* methyl or ethyl). Compounds of formula **VII** can also be synthesised from the piperazines **IV** and a compound of the formula $R_3\text{-}CH{=}CH\text{-}C(O)\text{-}X_1$. This alternative method of preparing Intermediates of formula **V** by acylating starting materials of formula **II** is useful also when Z is $CHOHCH_2$, if the OH group is previously protected by procedures which are well-known to those of ordinary skill in the art.

Schem 2.



Intermediates with formula V can be then reduced to intermediates of formula VI by the use of reducing agents which can convert the amido functionality into an amino group. Such reducing agents include complex metal hydrides, e.g. lithium aluminium hydride in diethyl ether or tetrahydrofuran, or a stable diborane complex such as borane-tetrahydrofuran or borane-dimethyl sulphide, or the like (*J. Org. Chem* **1982**, *47*, 1389) used in a solvent suitable for reducing reactions, e.g. tetrahydrofuran. When Z is $CH_2C(O)$, these reduction procedures can in any case be applied if the keto group is previously protected by procedures which are well-known to those of ordinary skill in the art.

Subsequent acylation of intermediates of formula VI with R-C(O)Cl or other acylating reagents following conventional procedures (see above) results in compounds of the invention of formula I. Intermediates of formula VI can also be acylated as their azaanion, which can be produced by treatment of intermediates of formula VI with hard bases (*e.g.* sodium hydride, butyl lithium, lithium bis(trimethylsilyl)amide) in a suitable solvent (*e.g.* tetrahydrofuran, toluene, dioxane, 1,2-dimethoxyethane, diglyme).

Another method to synthesise compounds of formula **I**, is depicted in Scheme 2 (above). Heteroaryl compounds with formula **II** (Y = halogen) are used to alkylate protected aminoalkylaldehydes or aminoalkylketones respectively of formula **VIII** (X = NH_2) to give the corresponding acetals or ketals of heteroarylaminoalkylaldehydes or heteroarylamino alkylketones **IX**. The reaction can be carried out in a polar aprotic solvent such as pyridine, DMF, toluene, etc., at temperatures between +40°C and +120°C, optionally in the presence of a base such as Et_3N.

An alternative reaction procedure to prepare intermediates of formula **IX** consists of alkylating heteroaryl compounds of formula **II** (Y=NH_2) with a protected compound of formula **VIII** (X=Br or other leaving group) by conventional procedures, or via an azaanion of compounds of formula **II**, obtained by the use of a strong base (e.g. n-butyl lithium, lithium diisopropylamide, lithium hexamethyldisilylamide, sodium hydride, sodium amide) in apolar or polar aprotic solvents (e.g. toluene, THF).

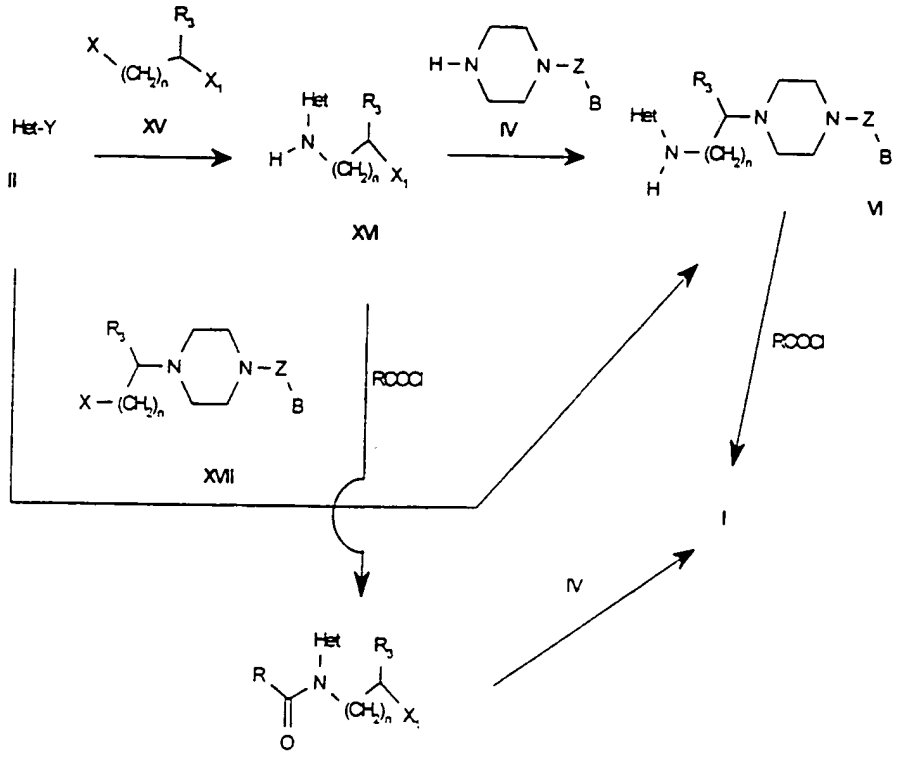
Intermediates of formula **IX** may be acylated with RCOCl to yield intermediates of formula **X** using the same acylation conditions described above (see Scheme 1). These intermediates are stable to normal laboratory conditions (i.e., light, humidity, temperature, etc.) and can be stored and deprotected by standard methods (e.g., acidic hydrolysis) just before their use in the following steps.

Carbonyl derivatives of formula **XI**, obtained from deprotection of compounds **X**, can be reacted with an appropriate N-substituted piperazine of formula **IV** under reductive amination conditions to give the compounds of the invention **I**. These reactions can be carried out in polar solvents such as methanol or ethanol, or chlorinated solvents such as dichloromethane or chloroform. Typically, alkali borohydrides such as $NaBH_4$, $NaBH_3CN$, and $NaBH(OAc)_3$ are employed as reducing agents. Additional, optional reaction components include acidic promoters such as acetic acid. The reactions proceed at temperatures between +10°C and 100°C. When compounds with formula **IV** (Z = CH_2CO) are used, the reduction of a ketone to an alcohol functionality can be achieved with $NaBH_4$ as a reducing agent.

Alternatively, carbonyl compounds of formula **XI** may be reacted with an appropriate N-protected piperazine of formula **XII**, using the same reductive conditions described above, to give intermediates with structure **XIII**. The piperazinyl derivatives of formula **XIV**, obtained by deprotection of compounds with formula **XIII** by standard methods, can be alkylated with B-Z-X reagents (X = leaving group) or B-Z-CHO reagents (by standard reductive amination, see above), except when Z is O and OCH_2, to give the compounds of the invention **I**.

An alternative method to synthesise the compounds of formula **I** is depicted in scheme 3.

Scheme 3.



The heteroarylamines of formula **II** (Y = NH_2) can be alkylated with an appropriate 1,w-disubstituted alkane of formula **XV** to give an intermediate with formula **XVI**. Reactions can be carried out in polar aprotic solvents such as N,N-dimethylformamide (DMF), tetrahydrofuran (THF), dioxane, acetone, acetonitrile or chlorinated solvents such as dichloromethane, or chloroform, at temperatures between 0°C and 120°C. The reactions are typically performed in the presence of a proton acceptor such as triethylamine (Et_3N), diisopropylethylamine, and the like, and optionally in the presence of potassium iodide.

In the intermediates of formula **XV**, X and X_1 can be chloro, bromo, iodo, alkyl- or arylsulphonyloxy.

Another approach to the synthesis of intermediates of formula **XVI** utilises compounds with formula **II** (Y = halogen, OTf) as starting materials. Het-Y is reacted with a suitable aminoalcohol of formula **XV** (X = NH_2, X_1 = OH or protected OH) by direct nucleophilic displacement performed by conventional procedures known to those skilled in the art, usually in the presence of bases (e.g. diisopropylethylamine, sodium carbonate, lithium

diisopropylamide, sodium *tert*-butoxide, etc.). One equivalent of excess reagent **XV** having X=NH_2 as proton acceptor is employed in the reaction, as described by G. Doleschall et al., Tetrahedron, **32**, 57-64 (1976). These alkylation reactions can be carried out in an aprotic polar solvent such as DMF, toluene, and the like, or in a protic one such as *n*-BuOH at temperatures between +40°C and +140°C. The nucleophilic displacement can be obtained also via arylation of an azaanion of **XV** (with protected OH) obtained by the use of hard bases (e.g. sodium hydride, butyl lithium, lithium bis(trimethylsilyl)amide) in a suitable solvent (e.g. tetrahydrofuran, toluene, dioxane, 1,2-dimethoxyethane, diglyme). The nucleophilic displacement of the aminoalcohol **XV** on Het-Y (Y = hal, Otf) can also be performed in the presence of a metal catalyst that can be chosen among a large variety e.g. copper metal, copper (I) iodide or bromide or oxide (*Tetrahedron*, **1984**, *40*, 1433), nickel catalysts (*J. Org. Chem.*, **1975**, *40*, 2267) palladium dichloride, palladium diacetate, palladium tetrakis, bis(diphenylphosphine) palladium dichloride, palladium dibenzylidene acetone, bis(diphenylphosphinoferrocene) palladium dichloride (*Synlett*, **1996**, 329; *J.Org. Chem.*, **1997**, *62*, 1568; **1997**, *62*, 1268; **1997**, *62*, 1264). The reactions can be performed at between room temperature and the reflux temperature of the solvent (e.g. dimethylacetamide, dimethylformamide, dioxane, acetonitrile, toluene, tetrahydrofuran) with or without a phosphine ligand (e.g. triphenylphosphine or tri-o-tolyl-phosphine or bis(diphenylphosphino)ferrocene or 2,2'-bis(diphenylphosphino)-1,1'-binaphthyl or other commercially available phosphine ligands).

Aminoalcohols **XVI** (X_1 = OH) are reacted with an halogenating agent such as $POCl_3$, $SOCl_2$, PCl_5 or PBr_3 to give intermediates of formula **XVI** (X_1 = Cl, Br). These reactions are typically carried out in an aprotic solvent such as chloroform, DMF, or pyridine at temperatures between +50°C and the reflux temperature of the solvent. These intermediates are used in the alkylation of an appropriate piperazine derivative with formula **IV**, to give intermediates of formula **VI**. These alkylations may be carried out in a chlorinated solvent such as dichloromethane, chloroform or 1,2-dichloroethane, or in a polar aprotic solvent such as DMF, THF, acetone, acetonitrile, *n*-butanol, etc., or in an apolar solvent such as toluene, benzene, *n*-heptane, and the like, at temperatures between 0 °C and 120 °C. The reaction mixture can, optionally, contain a proton acceptor such as Et_3N, 4-dimethylaminopyridine, potassium carbonate, caesium carbonate, and the like. The reaction can also optionally be performed in the presence of potassium iodide.

Intermediates with formula **VI** may be also obtained by the alkylation reaction of compounds of formula **II** (Y = NH_2) with derivatives of formula **XVII**, in which B, R_3, Z, X and n have the same meanings as defined above. Such reactions may be carried out at the melting temperature of the reactants without solvent, or in an aprotic solvent such as dichloromethane, chloroform, DMF, THF, acetone, acetonitrile, or a protic one such as *n*-butanol, and the like at temperatures between 0°C and +160°C. The reactions can.

optionally, be performed in the presence of a proton acceptor, such as Et_3N, potassium or caesium carbonate, 4-dimethylaminopyridine; another optional reactant is potassium iodide. Additionally, intermediates of formula **VI** may be prepared by nucleophilic displacement of a compound of formula **XVII** ($X = NH_2$) on a compound of formula **II** (Y = Hal, OTf) by the methods discussed above for the preparation of **XVI** ($X_1 = OH$).

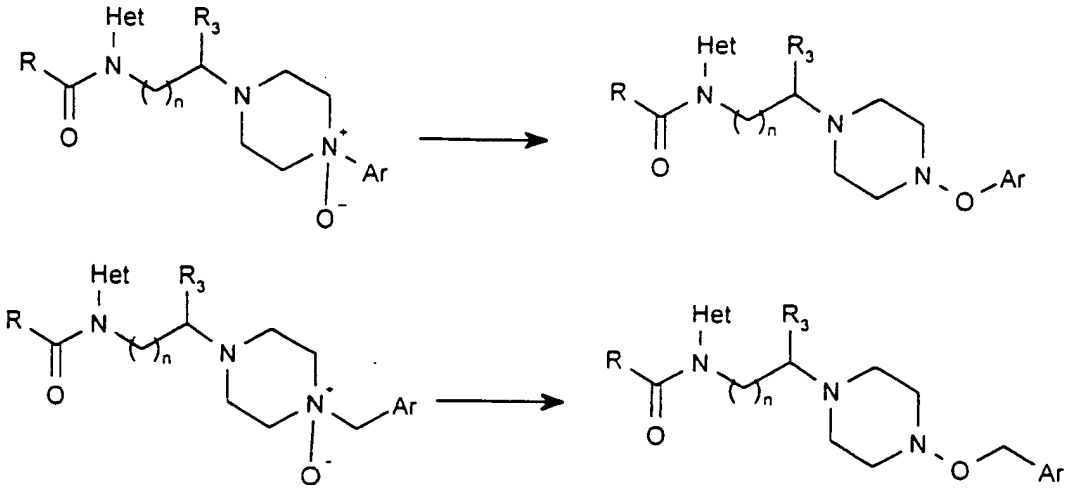
Intermediates of formula **VI** can be acylated with suitable acyl chlorides to give compounds of formula **I** as described above.

An additional procedure to afford compounds of formula **I** is acylation of compounds **XVI** by conventional procedure to give compounds **XVIII** (X_1 = leaving group), which can be finally reacted with the proper piperazine derivatives **IV**.

Intermediates with formulas **IV** and/or **VI** or **XVII** in which Z = $CH_2CH(OH)$ are obtained from the same intermediates in which Z = $CH_2C(O)$ by reduction. The reduction of carbonyl groups to alcohols may be carried out using metal hydrides such as $LiAlH_4$ (Rickborn J., J. Org. Chem. **35**, 1041 (1970)), $NaBH_4$, $Na(OAc)_3BH$ (Gribble N., Tetrahedron Lett. **24**, 4287 (1983)), $Zn(BH_4)_2$ (Chakraborty R., Tetrahedron Lett. **31**, 7663 (1990)) and the like as reducing agents. Diethyl ether, THF, methanol, ethanol, dioxane and solvent mixtures thereof that are suitable for use with strong bases and reducing agents may be used as reaction solvents, at temperatures between +10°C and the reflux temperature of the solvent.

Alternatively, these reductions may be carried out to achieve compounds of formula **I** where Z is $CH_2C(O)$ by employing lithium tri-*t*-butoxyaluminium hydride (Endy L., J. Org. Chem. **35**, 549 (1970)) or other selective reducing agents.

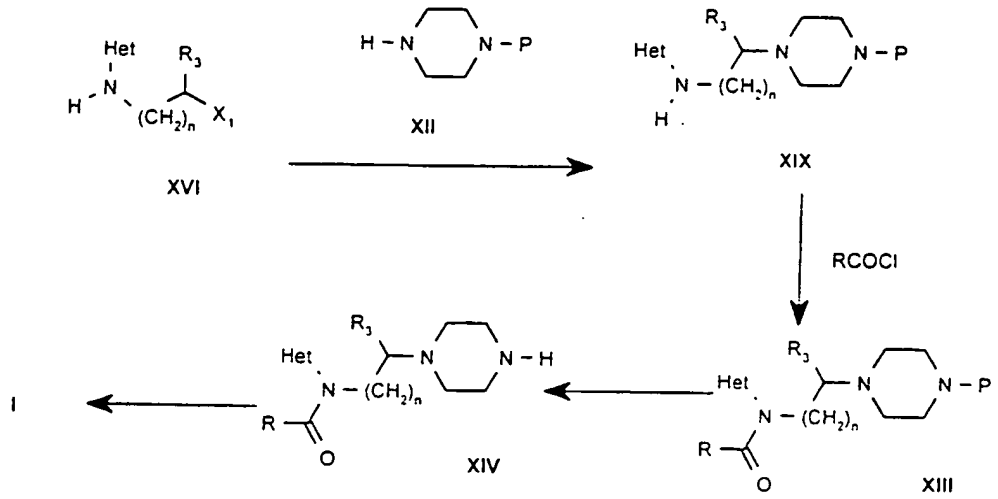
The compounds of formula **I** where Z is O or OCH_2 and B is aryl may be prepared from the corresponding N-oxide derivatives of compounds **I** in which Z is a bond or CH_2 by thermal rearrangement (Meisenheimer isomerization):



This reaction can be carried out in a polar aprotic solvent such as dioxane (Khuthier A.H. et al., J. Org. Chem. 52, 1710-1713 (1987) and references cited herein), at temperatures between 60°C and the reflux temperature of the solvent.

Another method to synthesise compounds of formula I is depicted in scheme 4, below.

Scheme 4.



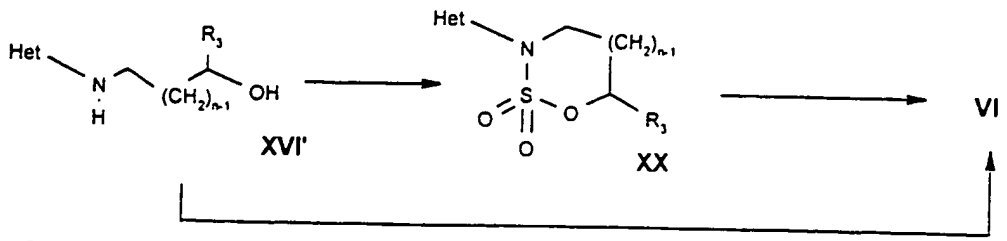
Intermediates of formula **XVI**, obtained as described in Scheme 3, may be used to alkylate mono-protected piperazines of formula **XII**, where P is *t*-butoxycarbonyl or benzyloxycarbonyl or other suitable protecting group, to afford intermediates with formula **XIX**. Several examples of protection and deprotection for various reactive groups can be found in: T. W. Greene, "Protective Groups in Organic Synthesis" Wiley Interscience (1991).

The reaction conditions for the preparation of intermediates of formula **XIX** are the same as described above in scheme 3.

Acylation of intermediates of formula **XIX** en route to the synthesis of intermediates with formula **XIII** (Scheme 2) can be performed under the same conditions employed for the synthesis of final compounds **I** in scheme 1.

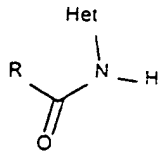
Another method for the synthesis of intermediates with formula **VI** consists of alkylating the appropriate arylpiperazines with intermediates of formula **XX**, whose synthesis is depicted in scheme 5:

Schem 5



where Het, n and R_3 have the same meanings as above and X_1 is OH.

Especially when Het is 2-pyridyl, but also in other cases, amino alcohol **XVI'** obtained as described in Scheme 3, can be converted into a reactive oxathiazolidine-2,2-dioxide **XX** (PCT/WO 95/33743). Intermediates of formulas **XVI'** or **XX** are reacted with a suitable piperazine derivative to afford intermediates of formula **VI**. These reactions are performed by conventional methods well known to those skilled in the art. Usually the condensation is carried out in an aprotic (*e.g.* acetonitrile, dimethylformamide, toluene, dioxane, tetrahydrofuran) or protic solvent (*e.g.* ethanol, n-butanol) or without any solvent, in the presence or absence of a base (*e.g.* triethylamine, diisopropylethylamine, pyridine, 4-dimethylaminopyridine, potassium carbonate) at a temperature between room temperature and 180°C.

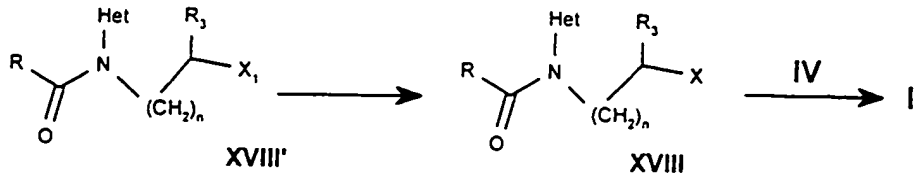


XXI

Another method for synthesising the compounds of the invention comprises alkylating an azaanion of amides of the formula **XXI**, above, wherein R and Het have the same meanings specified above.

The azaanion may be formed by using a base (e.g. sodium amide, butyl lithium, lithium diisopropylamide, sodium hydride, etc.) and the alkylating agent may be, for example, a compound of formula $X\text{-}(CH_2)_n\text{-}CH(R_3)\text{-}X_1$, where X_1 can be Br, Cl, OH or protected OH (e.g. O-tetrahydropyranyl) obtaining compounds of formula **XVIII** (X = leaving group), or **XVIII'** (X_1 = OH), as described above. As depicted in Scheme 6, compounds **XVIII'** can conveniently be converted into compounds **XVIII** through the already described halogenation procedure (preceded or not by a deprotection step).

Scheme 6



Compounds of formula **XVIII'** (X_1 = OH) can also be prepared by acylating compounds with formula **XVI** by conventional procedures, followed by alkaline hydrolysis of the esters obtained thereby.

A further method to obtain the compounds of the invention comprises alkylating the azaanion of amides **XXI** with a 1-(w-X-alkyl)-4-Z-B derivative of formula **XVII** (X = leaving group *e.g.* Br, Cl, I, p-toluenesulphonyloxy, methylsulphonyloxy).

EMBEDThe azaanion of amides **XXI** can be formed and alkylated by the use of a base (e.g. butyl lithium, sodium amide, sodium hydride, lithium diisopropyl amide, lithium *bis*(trimethylsilyl)amide or other known to those skilled in the art) in a proper aprotic solvent such as toluene, tetrahydrofuran, dimethoxyethane, dioxane, diglyme or others, at temperatures between -20°C and the reflux temperature of the solvent.

The compounds of the invention of formula Ia wherein R_1 is nitro can be easily converted into compounds of formula **I** where R_1 is amino, acylamino, or alkylsulfonylamino by conventional reaction procedures, such as by reduction of the nitro group via catalytic hydrogenation, transfer hydrogenation or well-known chemical methods to afford amino compounds with formula **I**, which can be then properly acylated or methylsulphonylated by known methods.

The following examples are provided merely to illustrate the invention and its advantages, and are not meant in any way to constitute a limitation on the scope of the invention. The invention is meant to encompass those obvious modifications of the compounds and methods described herein.

EXAMPLE 1

1-[*N*-cyclohexylcarbonyl-*N*-(2-pyridyl)-2-aminoethyl]-4-(2-trifluoromethoxyphenyl)-piperazine.

13.1 ml of a 2.5N solution of butyl lithium in hexane was added dropwise to a solution of 5.97 g 2-(2,2-dimethoxyethylamino)-pyridine [prepared as described by I. Kaye, in *J. Am. Chem. Soc.*, **73**, 5467 (1951)] in 40 ml tetrahydrofuran, stirred under a nitrogen atmosphere at 0°C. The cooling bath was then removed, and the mixture was stirred at room temperature for 1 hour, after which 4.46 ml cyclohexylcarbonyl chloride was added dropwise. Stirring was continued for 5.5 hours at room temperature, and then 2 ml of methanol was added. The solution was then evaporated to dryness *in vacuo*, yielding a

chestnut-coloured oil which was purified by flash chromatography (chloroform : ethyl acetate 7 : 3). The fractions containing the product were evaporated to dryness affording 8.3 g of N-(2,2-dimethoxyethyl)-N-(2-pyridyl)-cyclohexylcarboxamide (yield 78%); this substance was suitable for use in subsequent reactions without additional purification.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (dd, 1H, H6); 7.75 (dt, 1H, H4); 7.30 (d, 1H, H3); 7.20 (dt, 1H, H5); 4.65 (t, 1H, C<u>H</u>$(OCH_3)_2$); 3.90 (d, 2H, C<u>H_2</u>CH); 3.31 (s, 6H, 2 OCH_3); 2.20 - 2.43 (m, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

A solution of 0.6 g of N-(2,2-dimethoxyethyl)-N-(2-pyridyl)-cyclohexylcarboxamide and 0.02 g of hydroquinone in 10 ml of 2N hydrochloric acid was stirred, under a nitrogen atmosphere, for 30 minutes at a temperature of 80°C. The mixture was then cooled using a water bath and ice, and adjusted to pH 9 by addition of 5% aqueous sodium carbonate solution. The mixture was extracted with dichloromethane (2 x 10 ml); the extracts were combined, dried over anhydrous sodium sulphate and evaporated to dryness to give 68% yield of N-formylmethyl-N-(2-pyridyl)-cyclohexylcarboxamide, suitable for use in subsequent reactions without additional purification.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 9.66 (s, 1H, CHO); 8.50 (dd, 1H, H6); 7.79 (dt, 1H, H4); 7.20 - 7.38 (m, 2H, H3, H5); 4.52 (s, 2H, C<u>H_2</u>CHO); 2.37 - 2.55 (m, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

A mixture of 0.459 g of N-formylmethyl-N-(2-pyridyl)-cyclohexylcarboxamide, 0.445 g of 1-(2-trifluoromethoxyphenyl)-piperazine (EP 711757), 0.634 g of sodium triacetoxyborohydride, 0.23 ml of glacial acetic acid and 15 ml of 1,2-dichloroethane was stirred for 4 hours under a nitrogen atmosphere. The resultant solution was kept overnight at room temperature. The next day 10 ml of water was added and the solution was made alkaline by addition of 20% aqueous sodium carbonate solution. The phases were separated. The aqueous phase was extracted with 2 x 20 ml dichloromethane and the combined organic phases were dried over anhydrous sodium sulphate and evaporated to dryness *in vacuo*. The residue was purified by flash chromatography (ethyl acetate : petroleum ether, gradient 90:10 to 10:0). The solvents were evaporated off to give 0.44 g of the title compound (yield 51%).

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.52 (dd, 1H, pyridine H6); 7.74 (ddd, 1H, pyridine H4); 7.16 - 7.34 (m, 4H, pyridine H3 and H5, 2 aromatics of trifluoromethoxyphenyl); 6.33 - 7.02 (m, 2H, 2 aromatics of trifluoromethoxyphenyl); 3.97 (t, 2H, C(O)NC<u>H_2</u>CH_2); 2.85 - 3.05 (m, 4H, piperazine protons); 2.45 - 2.70 (m, 6H, piperazine protons, C(O)NCH_2C<u>H_2</u>); 2.10 - 2.32 (m, 1H, CHC(O)); 0.80 - 1.80 (2m, 10H, cyclohexyl protons).

EXAMPLE 2

<u>1-[*N*-cyclohexylcarbonyl-*N*-(2-pyridyl)-2-aminoethyl]-4-[2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine</u>

The title compound was prepared as described in the final step of Example 1, but using 1-[2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine (EP 748800) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The obtained residue was purified by flash chromatography (ethyl acetate : petroleum ether, gradient 90:10 to 10:0). Evaporation of the solvents afforded the title compound, yield 51%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.46 (dd, 1H, pyridine H6); 7.68 (ddd, 1H, pyridine H4); 7.06 - 7.28 (m, 2H, pyridine H3 and H5); 6.72 - 7.02 (m, 4H, aromatics of trifluoroethoxyphenyl); 4.30 (q, 2H, OCH_2CF_3); 3.91 (t, 2H, $C(O)NCH_2CH_2$); 2.80 - 3.05 (m, 4H, piperazine protons); 2.40 - 2.55 (m, 6H, piperazine protons, $C(O)NCH_2CH_2$); 2.05 - 2.25 (m, 1H, CHC(O)); 0.70 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 3

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-[5-chloro-2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine:

The title compound was prepared as described in the final step of Example 1, but using 1-[5-chloro-2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine (EP 748800) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The obtained residue was purified by flash chromatography (ethyl acetate : petroleum ether, gradient 90:10 to 10:0). Evaporation of the solvents afforded the title compound, yield 19.5%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.54 (dd, 1H, pyridine H6); 7.70 (ddd, 1H, pyridine H4); 7.15 - 7.33 (m, 2H, pyridine H3 and H5); 6.70 - 6.95 (m, 3H, aromatics of trifluoroethoxyphenyl); 4.32 (q, 2H, OCH_2CF_3); 3.97 (t, 2H, $C(O)NCH_2CH_2$); 2.85 - 3.10 (m, 4H, piperazine protons); 2.45 - 2.65 (m, 6H, piperazine protons, $C(O)NCH_2CH_2$); 2.10 - 2.32 (m, 1H, CHC(O)); 0.75 - 1.85 (2m, 10H, cyclohexyl protons).

EXAMPLE 4

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-cyanophenyl)-piperazine:

The title compound was prepared as described in the final step of Example 1, but using 1-(2-cyanophenyl)-piperazine [prepared as described by Martin et al, in *J. Med. Chem.*, **32**, 1052 (1996)] in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The obtained residue was purified by flash chromatography (ethyl acetate : petroleum ether, gradient 90:10 to 10:0). Evaporation of the solvents afforded the title compound, yield 19.5%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.55 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.53 - 7.75 (dd, 1H, H3 of cyanophenyl); 7.45 (dt, 1H, H5 of cyanophenyl); 7.20 - 7.30 (m, 2H, pyridine H3 and H5); 6.97 (t, 1H, H4 of cyanophenyl); 6.94 (d, 1H, H6 of cyanophenyl); 3.99 (t, 2H, $C(O)NCH_2CH_2$); 3.06 - 3.19 (m, 4H, piperazine protons); 2.55 - 2.70 (m, 6H, piperazine protons, $C(O)NCH_2CH_2$); 2.35 - 2.15 (m, 1H, CHC(O)); 0.80 - 1.80 (2m, 10H, cyclohexyl protons).

EXAMPLE 5

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-nitro-2-methoxyphenyl)-piperazine

The title compound was prepared as described in the final step of Example 1, but using 1-(2-methoxy-4-nitrophenyl)-piperazine (WO 95/17831) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The crude product was purified by flash chromatography (ethyl acetate : methanol, gradient 98:2 to 95:5). Evaporation of the solvents afforded the title compound. Yield 60%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.54 (dd, 1H, pyridine H6); 7.85 (dd, 1H, nitrophenyl H5); 7.75 (ddd, 1H, pyridine H4); 7.68 (d, 1H, nitrophenyl H3); 7.18 - 7.33 (m, 2H, pyridine H3 and H5); 6.85 (d, 1H, nitrophenyl H6); 3.97 (t, 2H, C(O)NC$\underline{H_2}$CH$_2$); 3.91 (s, 3H, OCH_3); 3.05 - 3.20 (m, 4H, piperazine protons); 2.55 - 2.70 (m, 6H, piperazine protons, C(O)NCH$_2$C$\underline{H_2}$); 2.15 - 2.35 (m, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 6

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-amino-2-methoxyphenyl)-piperazine

A mixture of 1.70 g of 1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-nitro-2-methoxyphenyl)-piperazine (prepared as described in Example 5), 18 ml of tetrahydrofuran, 18 ml of methanol, 50 mg of Raney Nickel and 1 ml of hydrazine hydrate was stirred for 2 hours at room temperature. A further 1 ml hydrazine hydrate was added, and the solution was stirred for 1 further hour at a temperature of 50°C. The catalyst was then filtered out, and the recovered solution was evaporated to dryness, affording the crude product. Purification by flash chromatography (ethyl acetate : 3N methanolic ammonia 95:5) and removal of the solvents by evaporation afforded 1.33 g of the title compound (yield 84%).

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.52 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.25 - 7.35 (m, 2H, pyridine H3 and H5); 6.75 (d, 1H, aminophenyl H6); 6.24 (d, 1H, aminophenyl H5); 6.21 (d, 1H, aminophenyl H3); 3.97 (t, 2H, C(O)NC$\underline{H_2}$CH$_2$); 3.78 (s, 3H, OCH_3); 3.00 - 4.00 (bs, 2H, NH_2); 2.78 - 3.00 (m, 4H, piperazine protons); 2.52 - 2.70 (m, 6H, piperazine protons, C(O)NCH$_2$C$\underline{H_2}$); 2.22 (tt, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 7

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-acetylamino-2-methoxyphenyl)-piperazine

A mixture of 0.35 g of 1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-amino-2-methoxyphenyl)-piperazine (prepared as described in Example 6), 5 ml of chloroform and 0.063 ml of acetyl chloride was stirred for 2.5 hours at room temperature. The mixture was then diluted with 50 ml of dichloromethane, washed with a saturated sodium carbonate solution and twice with water, dried over anhydrous sodium sulphate and evaporated to dryness. The crude product was purified by flash chromatography (ethyl acetate : 3N methanolic ammonia 95:5) and the recovered fraction was evaporated to dryness, affording 0.300 g of the title compound (yield 78%).

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.12 - 7.34 (m, 4H, pyridine H3 and H5, methoxyphenyl H6 and NH); 6.75 - 6.85 (m, 2H, methoxyphenyl H3 and H5); 4.00 (t, 2H, $C(O)NC\underline{H}_2CH_2$); 3.82 (s, 3H, OCH_3); 2.80 - 3.05 (m, 4H, piperazine protons); 2.50 - 2.71 (m, 6H, piperazine protons and $C(O)NCH_2C\underline{H}_2$); 2.05 - 2.10 (m, 1H, CHC(O)); 2.14 (s, 3H, CH_3CO); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 8

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-methylsulphonylamino-2-methoxyphenyl)-piperazine

This compound was prepared and purified by the method described in Example 7, but using methylsulphonyl chloride instead of acetyl chloride. Yield 75%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (d, 1H, pyridine H6); 7.55 (ddd, 1H, pyridine H4); 7.18 - 7.32 (m, 2H, pyridine H3 and H5); 6.70 - 6.90 (m, 3H, methoxyphenyl aromatics); 3.95 (t, 2H, $C(O)NC\underline{H}_2CH_2$); 3.82 (s, 3H, OCH_3); 2.80 - 3.05 (m, 7H, piperazine protons and CH_3S); 2.50 - 2.72 (m, 6H, piperazine protons and $C(O)NCH_2C\underline{H}_2$); 2.05 - 2.20 (m, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 9

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-carbamoylphenyl)-piperazine

The title compound was prepared as described in the final step of Example 1, but using 1-(2-carbamoylphenyl)-piperazine (WO 96/14846) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The crude product was purified by flash chromatography (dichloromethane : methanol 95:5). Evaporation of the solvents afforded the title compound. Yield 9%.

^{1}H-NMR ($CDCl_3$, δ): 9.35-9.55 (br, 1H, $CONH(\underline{H})$), 8.53 (dd, 1H, pyridine H6), 8.13 (dd, 1H, phenyl H6), 7.78 (dd, 1H, pyridine H4), 7.45 (dd, 1H, phenyl H4), 7.10-7.32 (m, 4H, remaining aromatics), 5.60-5.83 (br, 1H, $CON\underline{H}(H)$), 3.97 (t, 2H, $CONC\underline{H}_2CH_2$), 2.83-3.00 (m, 4H, piperazine protons), 2.50-2.75 (m, 6H, piperazine protons and $CONCH_2C\underline{H}_2$), 2.21 (tt, 1H, CHC(O)), 0.90-1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 10

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2,5-dichlorophenyl)-piperazine

The title compound was prepared as described in the final step of Example 1, but using 1-(2,5-dichlorophenyl)-piperazine (prepared as described by R. Ratouis et al, in *J. Med. Chem.* **1965**, *8*, 104-107) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The crude product was purified by flash chromatography (ethyl acetate : methanol 9:1). Evaporation of the solvents afforded the title compound as an amorphous ivory solid. Yield 45.6%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.85 (dd, 1H, pyridine H6); 8.08 (ddd, 1H, pyridine H4); 7.50 - 7.67 (m, 3H, pyridine H3 and H5, dichlorophenyl H3); 7.27 (s, 1H, dichlorophenyl H6); 7.22 (dd, 1H, dichlorophenyl H4); 4.22-4.50 (t, 2H, C(O)NC$\underline{H_2}$); 3.15-3.38 (m, 4H, piperazine protons); 2.80-3.01 (m, 6H, piperazine protons, C(O)NCH$_2$C$\underline{H_2}$); 2.45-2.65 (m, 1H, CHC(O)); 1.15-2.15 (m, 10H, cyclohexyl protons).

EXAMPLE 11

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-methoxy-4-pivaloylaminophenyl)-piperazine

This compound was prepared by the method described in Example 7, but using pivaloyl chloride instead of acetyl chloride. The obtained residue was purified by flash chromatography (ethyl acetate : 2.2N methanolic ammonia 95:5) affording the title compound (49%).

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (d, 1H, pyridine H6); 7.87 (ddd, 1H, pyridine H4); 7.48 (d, 1H, phenyl H3); 7.18-7.25 (m, 3H, pyridine H3 and H5 and NH); 6.70-6.90 (m, 2H, phenyl H5,6); 3.95-4.15 (m, 2H, C(O)NC$\underline{H_2}$CH$_3$); 3.85 (s, 3H, OCH_3); 2.45-3.30 (m, 10H, piperazine protons and C(O)NCH$_2$C$\underline{H_2}$); 2.25 (s, 1H, CHC(O)); 0.85 - 1.80 (m, 19H, cyclohexyl protons and $(CH_3)_3C$).

EXAMPLE 12

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-butanoylamino-2-methoxyphenyl)-piperazine

This compound was prepared and purified by the method described in Example 11, but using butanoyl chloride in place of pivaloyl chloride. Yield 53%.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (d, 1H, pyridine H6); 7.87 (ddd, 1H, pyridine H4); 7.40 (d, 1H, phenyl H3); 7.18 - 7.35 (m, 2H, pyridine H3 and H5); 7.10 (s, 1H, NH); 6.75 - 6.85 (bs, 2H, phenyl H5,6); 4.00 (s, 2H, C(O)NC$\underline{H_2}$CH$_2$); 3.85 (s, 3H, OCH_3); 2.85-3.10 (m, 4H, piperazine protons); 2.55-2.80 (m, 6H, piperazine protons and C(O)NCH$_2$C$\underline{H_2}$); 2.15-2.30 (m, 3H, CHC(O) and NHCOC$\underline{H_2}$); 0.85 - 1.80 (m, 15H, cyclohexyl protons and CH$_2$C$\underline{H_2}$).

EXAMPLE 13

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-bromo-5-methoxybenzyl)-piperazine

The title compound was prepared as described in the final step of Example 1, but using 1-(2-bromo-5-methoxybenzyl)-piperazine (AU 71773/96) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The crude product was purified by flash chromatography (ethyl acetate : methanol, gradient 100:0 to 100:3). Evaporation of the solvents afforded the title compound. Yield 52% as a thick yellow oil.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (dd, 1H, pyridine H6), 7.60 (ddd, 1H, pyridine H4), 7.39 (d, 1H, bromophenyl ring H3), 7.32-7.15 (m, 2H, pyridine H3 and H5), 7.15-6.95 (m, 1H, bromophenyl ring H6), 6.65 (dd, 1H, bromophenyl ring H4),4.05-3.88 (m, 2H, $CONCH_2CH_2$), 3.80 (s, 3H, OCH_3), 3.56 (s, 2H, benzylic CH_2), 2.80-2.32 (m, 10H, $CONCH_2CH_2$ and piperazine protons), 2.32-2.10 (m, 1H, CHC(O)), 1.82-0.82 (m, 10H, cyclohexyl protons).

EXAMPLE 14

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2,5-dichlorobenzyl)-piperazine methylsulphonate hemihydrate

2.01 g of 2,5-dichlorobenzyl chloride was added to a mixture of 1.94 g 1-ethoxycarbonylpiperazine, 3.45 g anhydrous potassium carbonate and 20 ml of dimethylformamide, stirred at room temperature under a nitrogen atmosphere. After stirring for a further 24 hours at room temperature, the reaction mixture was poured into 200 ml of water and extracted with ethyl acetate (3 x 100 ml). The combined organic phases were dried over anhydrous sodium sulphate and then evaporated to dryness *in vacuo*. The residue was purified by flash chromatography (petroleum ether : ethyl acetate 85:15). The solvents were then evaporated off to give 2 g of 1-(2,5-dichlorobenzyl)-4-ethoxycarbonylpiperazine (yield 63%).

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 7.50 (d, 1H, H6); 7.27 (d, 1H, H3); 7.15 (dd, 1H, H4); 4.13 (q, 2H, CH_3CH_2O); 3.58 (s, 2H, benzyl CH_2); 3.50 (m, 4H, piperazine protons); 2.47 (m, 4H, piperazine protons); 1.26 (t, 3H, CH_3CH_2O).

A solution of 13 g of 1-(2,5-dichlorobenzyl)-4-ethoxycarbonylpiperazine in 35 ml of 37% hydrochloric acid was stirred at reflux temperature for 40 hours. 30 ml of water and 30 ml of ethyl acetate were then added, and the pH was adjusted to 11 by addition of 35% sodium hydroxide. The organic phase was then separated, dried over anhydrous sodium sulphate and evaporated to dryness *in vacuo*. The residue was purified by flash chromatography (chloroform : methanol 7:3). 4.46 g of 1-(2,5-dichlorobenzyl)-piperazine (yield 50%) was obtained.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 7.50 (d, 1H, H6); 7.26 (d, 1H, H3); 7.14 (dd, 1H, H4); 3.55 (s, 2H, benzyl CH_2); 3.00 - 2.85 (m, 4H, piperazine protons); 2.55 - 2.48 (m, 4H, piperazine protons); 1.76 (s, 1H, NH).

0.049 g of sodium cyanoborohydride was added to a mixture comprising 0.164 g of N-formylmethyl-N-(2-pyridyl)-cyclohexylcarboxamide, 0.210 g of 1-(2,5-dichlorobenzyl)-piperazine and 4 ml methanol, continuously stirred under nitrogen at room temperature. The resulting mixture was then stirred for 13 hours at room temperature. It was then diluted with 10 ml of water, and extracted with ethyl acetate (3 x 10 ml). The combined organic phases were then dried over anhydrous sodium sulphate and evaporated to dryness *in vacuo*. The residue was purified by flash chromatography (chloroform : methanol 95:5). The recovered fractions were evaporated to dryness and the residue was taken up in 2 ml of ethyl acetate. 0.08 ml of 2M ethanolic methylsulphonic acid was added, followed by diethyl ether until the complete precipitation of the product. 0.071 g (yield 19%) of precipitate was recovered by vacuum filtration.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.51 (dd, 1H, pyridine H6); 7.83 (ddd, 1H, pyridine H4); 7.12 - 7.50 (m, 6H, pyridine H3 and H5, phenyl protons and NH^+); 4.10 - 4.25 (m, 2H, $C(O)NC\underline{H_2}CH_2$); 3.50 - 3.85 (m, 4H, piperazine protons); 3.15 - 3.40 (m, 2H, $C(O)CH_2C\underline{H_2}$); 2.75 - 3.15 (m, 6H, piperazine protons, benzyl CH_2); 2.74 (s, 3H, CH_3S); 2.33 - 2.18 (m, 1H, cyclohexyl H1); 0.80 - 1.80 (2m, 11H, cyclohexyl protons, 0.5 H_2O).

EXAMPLE 15

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzylpiperazine.

A mixture of 0.3 g of N-formylmethyl-N-(2-pyridyl)-cyclohexylcarboxamide, 0.2 ml of 1-benzylpiperazine, 0.14 ml of acetic acid, 0.38 g of sodium triacetoxyborohydride and 7 ml of dichloromethane was stirred at room temperature for 24 hours. The mixture was then diluted with water and made alkaline with 5% aqueous sodium hydrogen carbonate. The organic layer was separated, dried on anhydrous sodium sulphate and evaporated to dryness *in vacuo*. The crude product was purified by flash chromatography (dichloromethane : methanol 95:5) affording 0.03 g of the title compound.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.50 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.10 - 7.40 (m, 7H, pyridine H3 and H5, phenyl protons); 4.00 (t, 2H, $C(O)NC\underline{H_2}$); 3.60 (s, 2H, $PhCH_2$); 2.30 - 2.90 (m, 10H, piperazine protons, $C(O)NCH_2C\underline{H_2}$); 2.10-2.25 (m, 1H, CHC(O)); 0.80 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 16

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-phenethylpiperazine

This compound was prepared as described in Example 15, but using 1-phenethyl-piperazine (prepared as described in Beil. EIII/IV. 62) in place of 1-benzylpiperazine. The

crude was purified by flash chromatography (chloroform : methanol 97:3) affording 61% yield of the title product.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.51 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.10 - 7.20 (m, 7H, pyridine H3 and H5, phenyl protons); 3.96 (t, 2H, $C(O)NC\underline{H}_2$); 2.45-2.90 (m, 14H, $PhC\underline{H}_2C\underline{H}_2$, $CH_2C\underline{H}_2N$ and piperazine CH_2s); 2.20 (tt, 1H, CHC(O)); 0.85 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 17

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzoylpiperazine

This compound was prepared as described in Example 15, but using 1-benzoyl-piperazine (prepared as described by K.-R. Jacobi Chem. Ber. **1933**, 113-116) in place of 1-benzylpiperazine. The crude was purified by flash chromatography (chloroform : methanol 97.5:2.5) affording 54% yield of the title product.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.51 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.15 - 7.45 (m, 7H, pyridine H3 and H5, phenyl protons); 3.95 (t, 2H, $CONC\underline{H}_2CH_2N$) 3.45-3.80 (m, 2H, eq. H3 and H5 piperazine protons); 3.15-3.45 (m, 2H, ax. H3 and H5 piperazine protons); 2.30-2.65 (m, 6H, $CH_2C\underline{H}_2N$ and remaining piperazine CH_2s); 2.18 (tt, 1H, CHCO); 0.85 - 1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 18

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzoylmethylpiperazine hydrochloride.

This compound was prepared as described in the final step of Example 1, but using 1-benzoylmethyl-piperazine (prepared according to the procedure described in Beil. 23, V/2, 200) in place of 1-(2-trifluoromethoxyphenyl)-piperazine. The crude product was purified by flash chromatography (ethyl acetate -2N methanolic ammonia, gradient 97:3 to 95:5). The residue obtained by evaporation of the collected fractions was dissolved in ethyl acetate. An excess of 5N isopropanolic hydrogen chloride was added and the precipitate was collected by filtration to give the title compound, yield 18%. M.p. 225-228°C dec.

^{1}H-NMR 200 MHz (DMSO-d6, δ): 8.55 (dd, 1H, pyridine H6); 7.90-8.00 (m, 3H, pyridine H4, phenyl H2 and H6); 7.70 (dd, 1H, pyridine H3); 7.50-7.65 (m, 3H, phenyl H3, H4 and H5); 7.35-7.50 (ddd, 1H, pyridine H5); 5.30-6.80 (bs, 1H, NH^+); 5.05 (s, 2H, CH_2CO); 4.05 (t, 2H, CH_2NCO); 3.45-3.65 (m, 8H, piperazine protons); 3.30 (t, 2H, CH_2CH_2NCO); 2.20 (q, 1H, CHC(O)); 0.80-1.70 (m, 10H, cyclohexyl protons).

EXAMPLE 19

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(ß-hydroxy-phenethyl)-piperazine

0.158 g of sodium borohydride was added at 10°C to a solution of 0.18 g of 1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzoylmethylpiperazine (prepared as described in Example 18) in 4.2 ml of methanol. After stirring for 3 hours at room temperature and standing overnight, the solvent was removed and the residue was taken up in water and extracted with ethyl acetate. The crude, obtained after solvent evaporation, was purified by flash chromatography (ethyl acetate - 2N methanolic ammonia 97.5:2.5) to give 0.112 g (61%) of the title compound, m.p. 140-142°C.

^{1}H-NMR 200 MHz ($CDCl_3$, δ): 8.55 (dd, 1H, pyridine H6); 7.75 (ddd, 1H, pyridine H4); 7.20-7.40 (m, 7H, pyridine H3 and H5, phenyl CHs); 4.65-4.80 (dd, 1H, CHOH); 3.90-4.05 (m, 3H, OH, CH_2NCO); 2.30-2.80 (m, 12H, piperazine protons, CH_2CHOH, CH_2CH_2NCO); 2.15-2.30 (q, 1H, CHC(O)); 0.90-1.80 (m, 10H, cyclohexyl protons).

EXAMPLE 20

Effects on Volume-Induced Rhythmic Bladder Voiding Contractions in Anaesthetised Rats

A. Methods:

Female Sprague Dawley rats weighing 225-275 g (Crl: CDo BR, Charles River Italia) were used. The animals were housed with free access to food and water and were maintained on a forced 12 h alternating light-dark cycle at 22-24°C for at least one week, except during the experiment. The activity on the rhythmic bladder voiding contractions was evaluated according to the method of Dray (J. Pharmacol. Methods, 13:157, 1985), with some modifications as in Guarneri (Pharmacol. Res., 27:173, 1993). Briefly, rats were anaesthetised by subcutaneous injection of 1.25 g/kg (5 ml/kg) urethane, after which the urinary bladder was catheterised via the urethra using PE 50 polyethylene tubing filled with physiological saline. The catheter was tied in place with a ligature around the external urethral orifice and was connected with conventional pressure transducers (Statham P23 ID/P23 XL). The intravesical pressure was displayed continuously on a chart recorder (Battaglia Rangoni KV 135 with DCI/TI amplifier). The bladder was then filled via the recording catheter by incremental volumes of warm (37°C) saline until reflex bladder voiding contractions occurred (usually 0.8-1.5 ml). For intravenous (i.v.) injection of bioactive compounds, PE 50 polyethylene tubing filled with physiological saline was inserted into the jugular vein.

From the cystometrogram, the number of contractions recorded 15 min before (basal values) and after treatment, as well as the mean amplitude of these contractions (mean height of the peaks in mmHg) was evaluated.

Since most compounds produced an effect that was relatively rapid in onset and led to a complete cessation of bladder contractions, bioactivity was conveniently estimated by measuring the duration of bladder quiescence (i.e., the duration of time during which no contractions occurred). After cessation of the effect of drug injection, the height of the peaks was compared with that previously recorded after the intravenous administration of vehicle alone. The potency of the tested compounds (ED_{50} value: the extrapolated doses inducing 30% reduction of amplitude of the contractions in 50% of treated rats) was evaluated on a quantal basis by the method of Bliss (Bliss C.I., Quart. J. Pharm. Pharmacol. 11, 192-216, 1938).

B. Results

The rapid distension of the urinary bladder in urethane-anaesthetised rats produced a series of rhythmic bladder voiding contractions whose characteristics have been described (Maggi et al., Brain Res., 380:83, 1986; Maggi, et al., J. Pharmacol. Exp. Ther., 230:500, 1984). The frequency of these contractions is related to the sensory afferent arm of reflex micturition and to the integrity of the micturition centre, while their amplitude is a property of the efferent arm of the reflex. In this model system, compounds that act mainly on the CNS (such as morphine) cause a block in voiding contraction, whereas drugs that act at the level of the detrusor muscle, such as oxybutynin, lower the amplitude of the bladder contractions.

The results obtained with the tested compounds are shown in Table 1. All the compounds of the present invention produced better results than could be achieved with Compound A with regard to the disappearance time of bladder contractions. They also resulted in a longer time of suppression of contractions than flavoxate.

Oxybutynin only decreased the amplitude of the contractions in a dose-related manner, with an ED_{50} value (the extrapolated dose inducing a 30% reduction of amplitude of the contractions in 50% of treated rats) of 240 mg/kg. That is, oxybutynin does not cause cessation of bladder contractions. This amplitude-reduction effect characteristic of oxybutynin, which can potentially cause lower bladder contractility and the undesirable retention of residual urine in the bladder after micturition, is not a characteristic of the compounds of the invention.

TABLE 1

Effects on rhythmic bladder voiding contractions after intravenous administration.

Data represent mean values ± S.E. of the duration of bladder quiescence (disappearance time of contractions in min after i.v. administration of compound).

Compound	Dose (μg/kg)	Bladder Quiescence
Compound A	3	2.99 ± 0.63
	10	8.31 ± 1.28
	30	9.52 ± 0.82
	100	11.25 ± 1.09
	1000	6.30 ± 1.42
	3000	9.25 ± 2.77
Ex. 1	100	5.85 ± 1.87
	1000	16.25 ± 1.38
	3000	18.40 ± 4.12
Ex. 2	30	3.00 ± 0.91
	100	8.70 ± 2.88
	1000	27.28 ± 8.70
Ex. 3	10	3.15 ± 1.74
	100	6.15 ± 1.19
	1000	10.13 ± 0.59
	3000	14.42 ± 2.36
Ex. 6	10	6.75 ± 1.66
	100	9.82 ± 1.18
	1000	15.00 ± 3.11
Ex. 7	100	7.97 ± 1.78
	300	8.65 ± 1.81
	1000	20.63 ± 3.31

Table 1 continues on the next page

TABLE 1 (continued)

Effects on rhythmic bladder voiding contractions after intravenous administration.

Data represent mean values ± S.E. of the duration of bladder quiescence (disappearance time of contractions in min after i.v. administration of compound).

Compound	Dose (μg/kg)	Bladder Quiescence
Ex. 8	100	2.03 ± 0.44
	1000	7.20 ± 1.07
	3000	14.20 ± 2.57
Ex. 14	30	2.80 ± 1.31
	100	10.12 ± 1.98
	300	21.52 ± 4.66
Flavoxate	1000	3.04 ± 0.96
	3000	5.30 ± 1.00
	10000	8.25 ± 1.90
Oxybutynin	100	0.92 ± 0.15
	300	2.87 ± 1.12
	1000	2.80 ± 0.60
	3000	6.27 ± 2.90

EXAMPLE 21

Radioreceptor Binding to recombinant 5-HT_{1A} receptors

A. Methods:

Genomic clone G-21 coding for the human 5-HT_{1A}-serotoninergic receptor is stably transfected in a human cell line (HeLa). HeLa cells were grown as monolayers in Dulbecco's modified Eagle's medium (DMEM), supplemented with 10 % fetal calf serum and gentamicin (100 mg/ml), 5% CO_2 at 37°C. Cells were detached from the growth flash at 95% confluence by a cell scraper and were lysed in ice-cold 5 mM Tris and 5 mM EDTA buffer (pH 7.4). Homogenates were centrifuged at 40000 x g x 20 min and pellets were resuspended in a small volume of ice-cold 5 mM Tris 5 and 5 mM EDTA buffer (pH 7.4) and immediately frozen and stored at -70°C until use. On the day of experiment, cell membranes were resuspended in binding buffer: 50 mM Tris HCl (pH 7.4), 2.5 mM $MgCl_2$, 10mM pargiline (Fargin et al., Nature 335, 358-360, 1988). Membranes were

incubated in a final volume of 1 ml for 30 min at 30°C with 0.2 - 1 nM [^{3}H]8-OH-DPAT, in absence or presence of competing drugs; non-specific binding was determined in the presence of 10 mM 5-HT. The incubation was stopped by addition of ice-cold Tris-HCl buffer and rapid filtration through 0.2% polyethyleneimine pretreated Whatman GF/B or Schleicher & Schuell GF52 filters.

B. Results

As shown in Table 2, the compounds of the present invention have a high affinity for serotonergic 5-HT$_{1A}$ receptors. These results show that this receptor has a role in the action which the compounds of the invention exert on the bladder.

TABLE 2

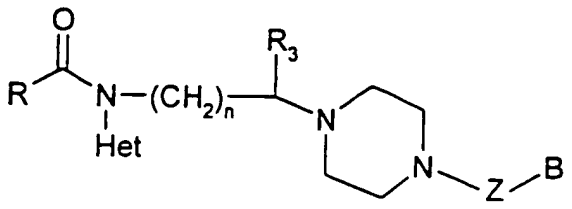
Binding affinity for the 5-HT$_{1A}$ receptor

Data are expressed as Ki (nM).

Compound	Ki (nM)
Compound A	0.39
Ex. 1	0.86
Ex. 2	0.89
Ex. 3	1.51
Ex. 4	0.31
Ex. 6	14.30
Ex. 7	8.15
Ex. 10	0.45
Ex. 14	0.12

CLAIMS

1. A compound of the formula



(I)

wherein

n is 1 or 2,

Het represents a monocyclic heteroaryl group,

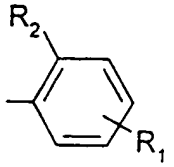
R represents a cycloalkyl or a monocyclic heteroaryl group,

R_3 represents a hydrogen atom or a lower alkyl group,

Z represents a bond or a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$, each of which is depicted with its left end being the end which attaches to the piperazine ring and its right end being the end which attaches to group B,

B represents a substituted or unsubstituted aryl or heteroaryl radical,

with the proviso that when Z represents a bond, B represents a substituted phenyl group of the formula



wherein

R_1 represents a hydrogen or halogen atom or an alkoxy, nitro, amino, acylamino or alkylsulphonylamino group, and

R_2 represents a halogen atom or an alkoxy, polyfluoroalkoxy, cyano or carbamoyl group, but

if R_1 does not represent an acylamino or alkylsulphonylamino group, then R_2 represents a polyfluoroalkoxy group;

or an enantiomer, diastereomer, N-oxide, crystalline form, hydrate, active metabolite or pharmaceutically acceptable salt of such a compound.

2. A compound according to claim 1 in which n is 1.

3. A compound according to claim 1 or claim 2 in which R represents a cyclohexyl group.

4. A compound according to any preceding claim in which Het represents a 2-pyridyl group.

5. A compound according to any preceding claim in which R_3 represents a hydrogen atom.

6. A compound according to any preceding claim in which Z represents a bond, R_1 represents a hydrogen or halogen atom or a nitro, amino, acylamino or alkylsulphonylamino group, and R_2 represents an alkoxy, trifluoroalkoxy, cyano or carbamoyl group.

7. A compound according to any preceding claim in which Z represents a bond and B represents a 2-trifluoromethoxyphenyl, 2-(2,2,2-trifluoroethoxy)-phenyl, 5-chloro-2-(2,2,2-trifluoroethoxy)-phenyl, 4-acetamido-2-methoxyphenyl, 2-methoxy-4-methylsulphonylaminophenyl, 2-methoxy-4-pivaloylaminophenyl or 4-butanoylamino-2-methoxyphenyl group.

8. A compound according to any of claim 1 to 5 in which Z represents a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$ or $-C(O)-$, each of which is depicted with its left end being the end which attaches to the piperazine ring and its right end being the end which attaches to group B.

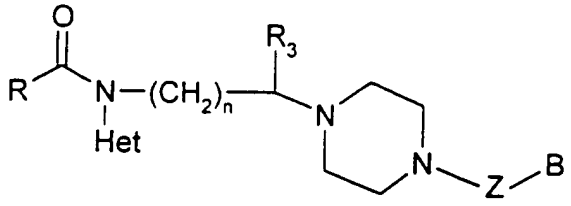
9. A compound according to any of claims 1 to 5 or 8 in which Z does not represent a valence bond and in which B represents a phenyl, 2,5-dichlorophenyl or 2-bromo-5-methoxyphenyl group.

10. Any one of the following compounds:
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-trifluoromethoxyphenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-[2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-[5-chloro-2-(2,2,2-trifluoroethoxy)-phenyl]-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-acetamido-2-methoxyphenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-methylsulphonylamino-2-methoxyphenyl)-piperazine.

1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-methoxy-4-pivaloylaminophenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-butanoylamino-2-methoxyphenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-bromo-5-methoxybenzyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2,5-dichlorobenzyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzylpiperazine.
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-phenethylpiperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzoylpiperazine.
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-benzoylmethyl-piperazine, and
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(ß-hydroxy-phenethyl)-piperazine;
or an enantiomer, diastereomer, N-oxide, crystalline form, hydrate, active metabolite or pharmaceutically acceptable salt of such a compound.

11. A pharmaceutical composition comprising a compound according to any preceding claim in admixture with a pharmaceutically acceptable diluent or carrier.

12. Use of a compound of the general formula I



(I)

wherein

n is 1 or 2,

Het represents a monocyclic heteroaryl group,

R represents a cycloalkyl or a monocyclic heteroaryl group,

R_3 represents a hydrogen atom or a lower alkyl group,

Z represents a bond or a group of the formula $-CH_2-$, $-CH_2CH_2-$, $-CH_2C(O)-$, $-CH_2CH(OH)-$, $-O-$, $-OCH_2-$ or $-C(O)-$, each of which is depicted with its left end being the end which attaches to the piperazine ring and its right end being the end which attaches to group B,

B represents a substituted or unsubstituted aryl or heteroaryl radical,

or an enantiomer, diastereomer, N-oxide, crystalline form, hydrate, active metabolite or pharmaceutically acceptable salt of such a compound, for the preparation of a medicament for the treatment of neuromuscular dysfunction of the lower urinary tract in a mammal.

13. Use according to claim 12 of a compound according to any of claims 1 to 10.

14. Use according to claim 12 of any one of the following compounds:
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-cyanophenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-nitro-2-methoxyphenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(4-amino-2-methoxyphenyl)-piperazine,
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2-carbamoylphenyl)-piperazine, and
1-[N-cyclohexylcarbonyl-N-(2-pyridyl)-2-aminoethyl]-4-(2,5-dichlorophenyl)-piperazine,
or of an enantiomer, diastereomer, N-oxide, crystalline form, hydrate, active metabolite or pharmaceutically acceptable salt of such a compound.

15. Use according to any of claims 12 to 14 for the preparation of a medicament which contains a pharmaceutically acceptable diluent or carrier.

16. Use according to any of claims 12 to 15 for the preparation of a medicament in a form suitable for oral administration.

17. Use according to claim 16 for the preparation of a medicament containing from 50 to 400 mg of the compound in unit dose form.

INTERNATIONAL SEARCH REPORT

Internationa. plication No
PCT/EP 98/04796

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C07D295/10 A61K31/495

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C07D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication where appropriate, of the relevant passages	Relevant to claim No.
Y	WO 95 04049 A (RECORDATI INDUSTRIA CHIMICA E FARMACEUTICA S.P.A ET AL.) 9 February 1995 see the whole document ---	1,11
Y	GB 2 255 337 A (JOHN WYETH & BROTHER LIMITED) 4 November 1992 cited in the application see the whole document ---	1,11
Y	WO 95 33743 A (JOHN WYETH & BROTHER LIMITED) 14 December 1995 cited in the application see the whole document ---	1,11
Y	WO 94 21610 A (JOHN WYETH & BROTHER LIMITED) 29 September 1994 see the whole document -----	1,11

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
4 January 1999	12/01/1999

Name and mailing address of the ISA	Authorized officer
European Patent Office, P B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Kyriakakou, G

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Information on patent family members

Internationa. plication No
PCT/EP 98/04796

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9504049 A	09-02-1995	IT MI931717 A	30-01-1995
		AU 680037 B	17-07-1997
		AU 7532394 A	28-02-1995
		CA 2168443 A	09-02-1995
		CN 1132508 A	02-10-1996
		EP 0711288 A	15-05-1996
		JP 9500883 T	28-01-1997
		NO 960371 A	29-03-1996
		NZ 271634 A	25-09-1996
		SG 46281 A	20-02-1998
		ZA 9405625 A	07-03-1995
GB 2255337 A	04-11-1992	AT 115566 T	15-12-1994
		AU 645681 B	20-01-1994
		AU 1524192 A	05-11-1992
		CA 2067929 A	03-11-1992
		CN 1098098 A	01-02-1995
		CS 9201344 A	14-10-1992
		DE 69200893 D	26-01-1995
		DE 69200893 T	13-04-1995
		DK 512755 T	30-01-1995
		EP 0512755 A	11-11-1992
		ES 2065133 T	01-02-1995
		FI 921942 A	03-11-1992
		HU 211148 B	30-10-1995
		IE 64634 B	23-08-1995
		IL 101722 A	14-05-1996
		JP 5170743 A	09-07-1993
		MX 9201991 A	01-11-1992
		ZA 9203081 A	28-10-1993
WO 9533743 A	14-12-1995	AU 680147 B	17-07-1997
		AU 2623095 A	04-01-1996
		BR 9507910 A	12-08-1997
		CA 2191871 A	14-12-1995
		CN 1149873 A	14-05-1997
		CZ 9603507 A	11-06-1997
		EP 0763031 A	19-03-1997
		FI 964815 A	02-12-1996
		HU 76466 A	29-09-1997
		JP 10500989 T	27-01-1998
		NZ 287498 A	28-10-1998
		SK 154196 A	06-08-1997
		US 5723464 A	03-03-1998
WO 9421610 A	29-09-1994	AT 170841 T	15-09-1998
		AU 6216394 A	11-10-1994
		DE 69413213 D	15-10-1998
		EP 0690845 A	10-01-1996
		JP 8508031 T	27-08-1996
		US 5693642 A	02-12-1997
		ZA 9401934 A	18-09-1995